SANTA CRUZ BIOTECHNOLOGY, INC.

CstF-64 (H-1): sc-398862

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The Power to Question

BACKGROUND

Polyadenylation of mRNA precursors is a two-step reaction that requires multiple protein factors. The first step, endonucleolytic cleavage of polyadenylation substrates, requires CstF (cleavage stimulation factor), a heterotrimer that is composed of three distinct subunits. CstF-64 contains an RNA binding domain and is responsible for the RNA binding activity of CstF. CstF-64 is expressed in all somatic cells and in pre- and postmeiotic, but not meiotic, germ cells. However, a large variant of CstF-64, called t CstF-64, is abundantly expressed in meiotic and postmeiotic cells in the testis and to a lesser extent in the brain, and promotes the germ cell pattern of polyadenylation. The gene encoding CstF-64 (designated CSTF2) maps to the X chromosome, whereas τ CstF-64 is encoded by an autosomal gene. The increase in CstF-64 concentration during B cell activation switches IgM heavy chain mRNA expression from membrane-bound to secreted forms, suggesting that CstF-64 plays a key role in regulating IgM heavy chain expression during B cell differentiation.

REFERENCES

1. Takagaki, Y., et al. 1990. A multisubunit factor, CstF, is required for polyadenylation of mammalian pre-mRNAs. Genes Dev. 4: 2112-2120.
2. Takagaki, Y., et al. 1996. The polyadenylation factor CstF-64 regulates alternative processing of IgM heavy chain pre-mRNA during B cell differentiation. Cell 87: 941-952.
3. Takagaki, Y. and Manley, J.L. 1998. Levels of polyadenylation factor CstF-64 control IgM heavy chain mRNA accumulation and other events associated with B cell differentiation. Mol. Cell 2: 761-771.

CHROMOSOMAL LOCATION

Genetic locus: CSTF2 (human) mapping to Xq22.1, CSTF2T (human) mapping to 10q21.1; Cstf2 (mouse) mapping to X E3, Cstf2f (mouse) mapping to 19 C1.

SOURCE

CstF-64 (H-1) is a mouse monoclonal antibody raised against amino acids 278-577 mapping at the C-terminus of CstF-64 of human origin.

PRODUCT

Each vial contains 200 µg IgM kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

APPLICATIONS

CstF-64 (H-1) is recommended for detection of CstF-64 and CstF-64T of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500), immunohistochemistry (including paraffin-embedded sections) (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

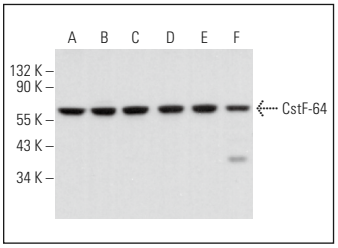
Molecular Weight of CstF-64: 64 kDa.

Positive Controls: HEK293 whole cell lysate: sc-45136, HeLa whole cell lysate: sc-2200 or Jurkat whole cell lysate: sc-2204.

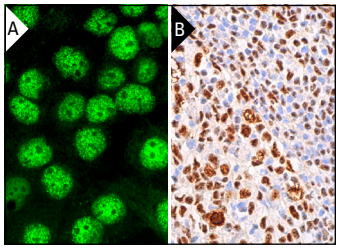
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein L-Agarose: sc-2336 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850. 4) Immunohistochemistry: use m-IgGκ BP-HRP: sc-516102 with DAB, 50X: sc-24982 and Immunohistomount: sc-45086, or Organo/Limonene Mount: sc-45087.

DATA



CstF-64 (H-1): sc-398862. Western blot analysis of CstF-64 expression in K-562 (**A**), HEK293 (**B**), HeLa (**C**), Jurkat (**D**), BJAB (**E**) and Hep G2 (**F**) whole cell lysates. Detection reagent used: m-IgGκ BP-HRP: sc-516102.



CstF-64 (H-1): sc-398862. Immunofluorescence staining of formalin-fixed A-431 cells showing nuclear localization (**A**). Immunoperoxidase staining of formalin fixed, paraffin-embedded human tonsil tissue showing nuclear staining of cells in germinal center, cells in non-germinal center and squamous epithelial cells (**B**).

SELECT PRODUCT CITATIONS

1. Gong, Q., et al. 2018. Regulation of Kv11.1 potassium channel C-terminal isoform expression by the RNA-binding proteins HuR and HuD. J. Biol. Chem. 293: 19624-19632.
2. Deka, K. and Saha, S. 2021. Heat stress induced arginylation of HuR promotes alternative polyadenylation of Hsp70.3 by regulating HuR stability and RNA binding. Cell Death Differ. 28: 730-747.
3. Mukherjee, S., et al. 2023. Macrophage differentiation is marked by increased abundance of the mRNA 3' end processing machinery, altered poly(A) site usage, and sensitivity to the level of CstF64. Front. Immunol. 14: 1091403.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.